ESI

EnviroSystems, Inc.
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603-926-3345

June 2, 2015

Mr. Aram Varjabedian
Woodard & Curran
Hull Water Pollution Control Facility
1111 Nantasket Avenue
Hull, Massachusetts 02045

Dear Mr. Varjabedian:

Enclosed, please find a copy of our report presenting the results of a toxicity test completed using an effluent sample collected from the Hull, Massachusetts Water Pollution Control Facility during the May 2015 sampling period. Acute toxicity was evaluated using the inland silverside minnow, *Menidia beryllina*.

Please do not hesitate to call me, Kirk Cram or Petra Karbe should you have any questions regarding the report.

Sincerely,

EnviroSystems, Incorporated

Kenneth A. Simon
Technical Director

Enclosure
WET Test Report Certification
Report Number 25910-15-05
One (1) copy + email

WHOLE EFFLUENT TOXICITY TEST REPORT CERTIFICATION

Permittee Certification

I certify under penalty of law that this document and all attachments were prepared under my direction or supervision in accordance with a system designed to assure that qualified personnel properly gather and evaluate the information submitted. Based on my inquiry of the person or persons who manage the system, or those persons who manage the system, or those persons directly responsible for gathering the information, the information submitted is, to the best of my knowledge and belief, true, accurate, and complete. I am aware that there are significant penalties for submitting false information, including the possibility of fine and imprisonment for knowing violations.

Executed on: _____

_____

Authorized Signature

_____

Print or Type Name

Hull Permanent Sewer Commission

Print or Type the Permittee's Name

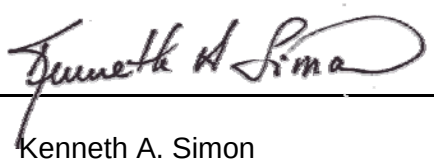
MA0101231

Type or Print the NPDES Permit No.

WHOLE EFFLUENT TOXICITY TEST REPORT CERTIFICATION (Bioassay Laboratory)

I certify under penalty of law that this document and all attachments were prepared under my direction or supervision in accordance with a system designed to assure that qualified personnel properly gather and evaluate the information submitted. Based on my inquiry of the person or persons who manage the system, or those persons who manage the system, or those persons directly responsible for gathering the information, the information submitted is, to the best of my knowledge and belief, true, accurate, and complete. I am aware that there are significant penalties for submitting false information, including the possibility of fine and imprisonment for knowing violations.

Executed on: June 2, 2015



Kenneth A. Simon
Technical Director - EnviroSystems, Inc.

TOXICOLOGICAL EVALUATION
OF A TREATED MUNICIPAL EFFLUENT
BIOMONITORING SUPPORT FOR A NPDES PERMIT:
May 2015

Hull Water Pollution Control Facility
Hull, Massachusetts
NPDES Permit Number MA0101231

Prepared For:

Woodard & Curran
Hull Water Pollution Control Facility
1111 Nantasket Avenue
Hull, Massachusetts 02045

Prepared By:

EnviroSystems, Incorporated
One Lafayette Road
Hampton, New Hampshire 03842

May 2015
Reference Number Hull25910-15-05

STUDY NUMBER 25910

EXECUTIVE SUMMARY

The following summarizes the results of an acute exposure bioassay completed during May 2015 in support of the NPDES biomonitoring requirements of the Hull, Massachusetts Water Pollution Control Facility, operated by Woodard & Curran. The 48 hour acute definitive assay was completed using the inland silverside minnow, *Menidia beryllina*.

M. beryllina were 8 days old at the start of the test. Dilution water was receiving water collected from Massachusetts Bay at a point away from the discharge. Samples were received under chain of custody in good order. All sample receipt, test conditions and control endpoints were within protocol specifications, except where otherwise noted.

The results presented in this report relate only to the samples described on the chain(s) of custody and sample receipt log(s), and are intended to be used only by the submitter. Results from the acute exposure assay and their relationship to permit limits are summarized in the following matrix.

Acute Toxicity Evaluation

Species	Exposure	LC-50	A-NOEC	Permit Limit (LC-50)	Effluent Meets Permit Limit	Assay Meets Protocol Limits
Menidia beryllina [a,b]	48 Hours	>100%	100% [c]	≥100%	Yes	Yes

COMMENTS:

[a] Twelve fish were added to replicate D of the laboratory water control at the start of the assay, therefore 12 organisms were used in the statistical analysis from the start of the assay.

[b] Replicate A of the 100% test concentration was removed from statistical analyses as all organisms were accidentally lost on test day 2.

[c] The statistical analysis for minnow growth was completed using the Dunn/Bonferroni and the linear interpolation (IC-25) tests, rather than following the standard US EPA decision tree, because of the number of replicates selected for the analysis at the specified alpha level.

TOXICOLOGICAL EVALUATION
OF A TREATED MUNICIPAL EFFLUENT
BIOMONITORING SUPPORT FOR A NPDES PERMIT:
May 2015

Hull Water Pollution Control Facility
Hull, Massachusetts
NPDES Permit Number MA0101231

1.0 INTRODUCTION

This report presents the results of an acute toxicity test completed on a composite effluent sample collected from the Hull, Massachusetts Water Pollution Control Facility (Hull WPCF), operated by Woodard & Curran. Testing was based on programs and protocols developed by the US EPA (2002) with exceptions as noted by US EPA Region I (2012) and involved conducting a 48 hour static acute toxicity test with the inland silverside minnow: *Menidia beryllina*. Testing was performed at EnviroSystems, Incorporated (ESI), Hampton, New Hampshire in accordance with the provisions of TNI Standards (2009).

Acute toxicity tests involve preparing a series of concentrations by diluting effluent with control water. Groups of test animals are exposed to each effluent concentration and a control for a specified period. In acute tests, mortality data for each concentration are used to calculate (by regression) the median lethal concentration, or LC-50, defined as the effluent concentration that kills half of the test animals. Samples with high LC-50 values are less likely to cause significant environmental impacts. The no-effect concentration is also determined to provide information about the level of effluent which would have minimal acute effects in the environment. This Acute No Observed Effect Concentration (A-NOEC) is defined as the highest tested effluent concentration that causes no significant mortality.

2.0 MATERIALS AND METHODS

2.1 General Methods

Toxicological and analytical protocols used in this program follow procedures primarily designed to provide standard approaches for the evaluation of toxicological effects of discharges on aquatic organisms (US EPA 2002), and for the analysis of water samples (APHA 2012). See Section 4.0 for a list of references.

2.2 Test Species

When necessary, *M. beryllina* were acclimated to approximate test conditions prior to use in the assay. Test organisms were transferred to test chambers using an inverted glass pipet, minimizing the amount of water added to test solutions. Twenty control fish were weighed during the test to confirm loading rates. The loading rate was below the maximum 0.4 g/L recommended for assays conducted at 25°C. Fish weights and loading calculations are included in the data appendix.

2.3 Effluent, Receiving Water, and Laboratory Water

Effluent and receiving water collection information is provided in Table 1. Samples were received at 0-6°C as per 40 CFR §136.3 unless otherwise noted, stored at 4±2°C and warmed to 25±1°C prior to preparing test solutions. Effluent used in the *M. beryllina* assay was salinity adjusted to 25±2 ppt using artificial sea salts according to protocol (US EPA 2002). Laboratory water was collected from the Hampton/Seabrook Estuary. This water is classified as SA-1 and has been used to culture marine test organisms since 1981.

Total residual chlorine (TRC) was measured by amperometric titration (MDL 0.02 mg/L) in both the effluent and diluent samples. If chlorine was present, the sample was dechlorinated using sodium thiosulfate. A control treatment using laboratory water adjusted with an equal amount of sodium thiosulfate to dechlorinate the sample was run concurrently. Data for the sodium thiosulfate laboratory control can be found in Appendix A.

2.4 Acute Toxicity Test

The 48 hour static acute toxicity test was conducted at 25±1°C with a photoperiod of 16:8 hours light:dark. Test chambers were 250 mL glass beakers containing 200 mL test solution in each of 4 replicates with 10 organisms/replicate. Replicates were not randomized during testing, rather organisms were added randomly at test initiation by replicate across test solutions in an alternating fashion (alternating allocation). Test concentrations for the assay were 100% (undiluted), 50%, 25%, 12.5%, and 6.25% effluent. Survival and dissolved oxygen were recorded daily in all replicates. Specific conductivity, salinity, temperature, and pH were measured daily in one replicate of each test treatment.

2.5 Data Analysis

When applicable, statistical analysis of acute exposure data was completed using CETIS™ v1.8.6.6, Comprehensive Environmental Toxicity Information System, software. The program computes acute exposure endpoints based on US EPA decision tree guidelines specified in individual test methods. If survival in the highest test concentration is >50%, the LC-50 is obtained by direct observation of the raw data. As needed, the A-NOEC is determined as the highest test concentration that caused no significant mortality.

2.6 Quality Control

As part of the laboratory quality control program, standard reference toxicant assays are completed on a regular basis for each test species. These results provide relative health and response data while allowing for comparison with historic data sets. See Table 2 for details.

3.0 RESULTS AND DISCUSSION

Results of the acute exposure bioassay completed using the inland silverside minnow are summarized in Table 3. Effluent and dilution water characteristics are presented in Table 4. US EPA Region I toxicity test summary sheet can be found after the tables. Support data, including copies of the laboratory bench sheets, are included in Appendix A.

Minimum test acceptability criteria require ≥90% survival in the control concentrations. Achievement of these results indicate that healthy test organisms were used and that the dilution water had no significant adverse impact on the outcome of the assay. See the Executive Summary and Table 3 for test acceptability.

4.0 LITERATURE CITED

40 CFR §136.3. *Code of Federal Regulations* (CFR), Protection of the Environment (Title 40), Guidelines Establishing Test Procedures for the Analysis of Pollutants (Part 136), Identification of Test Procedures (sub-part 3), Table II-Required Containers, Preservation Techniques, and Holding Times.

APHA. 2012. *Standard Methods for the Examination of Water and Wastewater*, 22nd Edition. Washington D.C.

The NELAC Institute (TNI). 2009. *Environmental Laboratory Sector, Volume 1: Management and Technical Requirements for Laboratories Performing Environmental Analysis (TNI Standard)*. EL-V1-2009.

US EPA. 2002. *Methods for Measuring the Acute Toxicity of Effluents to Freshwater and Marine Organisms*. Fifth Edition. EPA-821-R-02-012.

US EPA Region I. 2012. *Marine Acute Toxicity Test Procedure and Protocol*. US EPA Region I Office, Boston, Massachusetts. July 2012.

TABLE 1. Summary of Sample Collection Information.
Hull WPCF Effluent Biomonitoring Program. May 2015.

Sample Description	Type	Collection Date	Collection Time	Receipt Date	Receipt Time	Arrival Temp °C
Effluent	Comp	05/12-13/15	0800-0800	05/13/15	1100	4
Receiving Water	Grab	05/13/15	0700	05/13/15	1100	4

TABLE 2. Summary of Reference Toxicant Data.
Hull WPCF Effluent Biomonitoring Program. May 2015.

Date	Endpoint		Value	Historic Mean/ Central Tendency	Acceptable Range	Reference Toxicant
M. beryllina						
05/20/15	Survival	48Hr LC-50	7.0	6.3	4.1 - 8.5	SDS (mg/L)

Means and Acceptable Ranges based on the most recent 20 reference toxicant assays

TABLE 3. Summary of Acute Evaluation Results.
Hull WPCF Effluent Biomonitoring Program. May 2015.

Percent Survival

Species	Exposure	Lab	RW	6.25%	12.5%	25%	50%	100%
M. beryllina	48 hours	90.8% [a]	85%	87.5%	100%	97.5%	100%	93.3% [b]

LC-50 and A-NOEC Results

Species	Exposure	Spearman-Kärber	Probit	Direct Observation	A-NOEC
M. beryllina	48 Hours	NC	NC	>100%	100% [c]

COMMENTS:
RW = Receiving Water; used as diluent.
NC = Not Calculated.
[a] Twelve fish were added to replicate D of the laboratory water control at the start of the assay, therefore 12 organisms were used in the statistical analysis from the start of the assay.
[b] Replicate A of the 100% test concentration was removed from statistical analyses as all organisms were accidentally lost on test day 2.
[c] The statistical analysis for minnow growth was completed using the Dunn/Bonferroni and the linear interpolation (IC-25) tests, rather than following the standard US EPA decision tree, because of the number of replicates selected for the analysis at the specified alpha level.

TABLE 4. Summary of Effluent and Diluent Characteristics.
Hull WPCF Effluent Biomonitoring Program. May 2015.

PARAMETER	UNIT	EFFLUENT	RECEIVING WATER
Specific Conductivity - As Received	µmhos/cm	10350	45800
pH - As Received	SU	7.10	7.84
Salinity - As Received	ppt	6	29
Total Residual Chlorine	mg/L	<0.02	<0.02
Total Solids	mg/L	5000	35000
Total Suspended Solids	mg/L	23	6.3
Ammonia as N	mg/L	16	<0.1
Total Organic Carbon	mg/L	11	0.7
Aluminum, total	mg/L	0.09	0.099
Cadmium, total	mg/L	<0.0005	<0.0005
Chromium, total	mg/L	<0.002	<0.002
Copper, total	mg/L	0.37	0.006
Lead, total	mg/L	0.001	<0.0005
Nickel, total	mg/L	0.003	<0.002
Zinc, total	mg/L	0.14	0.007

COMMENTS:
Additional water quality and analytical support chemistry data are available in Appendix A.

TOXICITY TEST SUMMARY SHEET

FACILITY NAME: Hull WPCF
NPDES PERMIT NO.: MA0101231
TEST START DATE: 05/14/15
TEST END DATE: 05/16/15

TEST TYPE	TEST SPECIES	SAMPLE TYPE	SAMPLE METHOD
X Acute	___ *Pimephales promelas*	___ Prechlorinated	___ Grab
___ Chronic	___ *Ceriodaphnia dubia*	___ Dechlorinated	X Composite
___ Modified Chronic (Reporting Acute Values)	___ *Daphnia pulex*	___ Chlorine Spiked in Lab	___ Flow-thru
	___ *Americamysis bahia*	___ Chlorinated on Site	___ Other
	___ *Cyprinodon variegatus*	___ Unchlorinated	
___ 24 Hour Screen	X *Menidia beryllina*	X No Detectable Chlorine Upon Receipt	
	___ *Arbacia punctulata*	___ Dechlorinated at lab	

DILUTION WATER:

X Receiving water collected at a point upstream or away from the discharge, free from toxicity or other sources of contamination; Receiving Water Name: Massachusetts Bay

___ Alternate surface water of known quality and hardness, to generally reflect the characteristics of the receiving water; Receiving Water Name: _____

___ Synthetic water prepared using either Millipore Milli-Q or equivalent deionized water and reagent grade chemicals; or deionized water combined with mineral water.

___ Artificial sea salts mixed with deionized water

___ Deionized water and hypersaline brine

___ Other

EFFLUENT SAMPLING DATES: 05/12-13/15 _____ _____ _____

EFFLUENT CONCENTRATIONS TESTED (%): 6.25; 12.5; 25.0; 50.0; 100

Permit Limit Concentration: ≥100 %

Was the effluent salinity adjusted? Yes If yes, to what level? 26 ppt

REFERENCE TOXICANT TEST DATE: 05/20/15 LC-50: 7.0 mg/L Sodium Dodecyl Sulfate

PERMIT LIMITS AND TEST RESULTS

Test Acceptability Criteria

Mean Control Survival: 85%

LIMITS		**RESULTS**	
LC-50:	≥100 %	LC-50	>100 %
		Upper Limit:	- %
A-NOEC:	- %	Lower Limit:	- %
		Method:	Direct Observation
C-NOEC:	- %	A-NOEC:	100 %
		C-NOEC:	- %
		C-LOEC:	-
IC- ___	- %	IC- 25	>100 %

APPENDIX A

DATA SHEETS

STATISTICAL SUPPORT

METHODS USED IN NPDES PERMIT BIOMONITORING TESTING

Parameter	Method
Acute Exposure Bioassays:	
Ceriodaphnia dubia	EPA-821-R-02-012 2002.0
Daphnia pulex	EPA-821-R-02-012 2021.0
Pimephales promelas	EPA-821-R-02-012 2000.0
Americamysis bahia	EPA-821-R-02-012 2007.0
Menidia beryllina	EPA-821-R-02-012 2006.0
Cyprinodon variegatus	EPA-821-R-02-012 2004.0
Chronic Exposure Bioassays:	
Ceriodaphnia dubia	EPA-821-R-02-013 1002.0
Pimephales promelas	EPA-821-R-02-013 1000.0
Cyprinodon variegatus	EPA-821-R-02-014 1004.0
Menidia beryllina	EPA-821-R-02-014 1006.0
Arbacia punctulata	EPA-821-R-02-014 1008.0
Champia parvula	EPA-821-R-02-014 1009.0
Trace Metals:	
Trace Metals	EPA 200.8/SW 6020, EPA 245.7
Hardness	Standard Methods 22nd Edition - Method 2340 B
Wet Chemistries:	
Alkalinity	EPA 310.2
Chlorine, Residual	Standard Methods 22nd Edition - Method 4500-Cl D
Total Organic Carbon	Standard Methods 22nd Edition - Method 5310 C
Specific Conductance	Standard Methods 22nd Edition - Method 2510 B
Nitrogen - Ammonia	Standard Methods 22nd Edition - Method 4500-NH_3 G
pH	Standard Methods 22nd Edition - Method 4500-H+ B
Solids, Total (TS)	Standard Methods 22nd Edition - Method 2540 B
Solids, Total Dissolved (TDS)	Standard Methods 22nd Edition - Method 2540 C
Solids, Total Suspended (TSS)	Standard Methods 22nd Edition - Method 2540 D
Dissolved Oxygen	Standard Methods 22nd Edition - Method 4500-O G

Please visit our web site at www.envirosystems.com for a copy of our accreditations and state certifications.

ACUTE BIOASSAY DATA SUMMARY

STUDY: 25910	Brine Shrimp: A- 3714	"AS RECEIVED" EFFLUENT AND DILUENT CHEMISTRIES							
CLIENT: United Water	TEST ORGANISM: *M. beryllina*		T. Metals	TOC	AMM	TS/TSS	pH	S/C	SALINITY	TRC
SAMPLE: Hull WWTF Effluent	ORGANISM SUPPLIER / BATCH / AGE: *See Organism Culture Sheet*	EFF	001	003	004	005	7.10	10350	5.8	<0.02
DILUENT: Receiving Water		DIL	007	008	009	010	7.84	45800	29.4	<0.02

SALINITY ADJUSTMENT RECORD : 4000 ML EFFLUENT + 88 G SEA SALTS (A- 3700) = 100% ACTUAL PERCENTAGE (EH 05/13)
8000 mL Diluent + 2000 ml ~~Sea Salts~~ DI H2O (EH 05/13) = 80% ~~100%~~ Actual Percentage

CONC	REP	SURVIVAL 0	SURVIVAL 24	SURVIVAL 48	DO (mg/L) 0	DO (mg/L) 24	DO (mg/L) 48	pH (SU) 0	pH (SU) 24	pH (SU) 48	TEMP (°C) 0	TEMP (°C) 24	TEMP (°C) 48	S/C (µmhos/cm) 0	S/C (µmhos/cm) 24	S/C (µmhos/cm) 48	SALINITY (ppt) 0	SALINITY (ppt) 24	SALINITY (ppt) 48
LAB	A	10	10	10	7.6	5.7	5.6	8.03	7.83	7.83	22	24	24	39150	38220	39030	25	24	25
	B	10	10	10	7.6	5.9	5.9												
	C	10	8	8	7.6	5.9	5.9												
	D	10	10 (E10)	10	7.6	6.2	5.9												
RW	A	10	10	10	7.5	5.7	6.0	7.98	7.79	7.81	24	24	24	37920	38340	38510	24	25	24
	B	10	8	8	7.5	5.9	6.0												
	C	10	9	8	7.5	5.9	5.9												
	D	10	9	8	7.5	5.9	5.9												
6.25%	A	10	10	10	7.7	6.0	5.8	7.96	7.84	7.85	24	24	24	38440	38570	38860	24	25	25
	B	10	10	10	7.7	6.0	6.0												
	C	10	6	5	7.7	6.0	5.9												
	D	10	10	10	7.7	5.9	5.9												
12.5%	A	10	10	10	7.6	5.5	5.9	7.93	7.83	7.85	24	24	24	38690	38370	39160	25	25	25
	B	10	10	10	7.6	5.7	5.9												
	C	10	10	10	7.6	5.6	5.9												
	D	10	10	10	7.6	5.7	5.9												
DATE		5/14/15	5/15/15	5/16/15	05/14/15	5/15/15	5/16/15												
TIME		1430	1550	1345	1230	1400	1215												
INITIALS		BL	NP	BG	EH	NP	BL												

(E10) NP 5/15/15 2 dead, removed 10 left.

ACUTE BIOASSAY DATA SUMMARY

STUDY: 25910	**Brine Shrimp: A-** 3714	
CLIENT: United Water	**TEST ORGANISM:** *M. beryllina*	
SAMPLE: Hull WWTF Effluent	**ORGANISM SUPPLIER / BATCH / AGE:** *See Organism Culture Sheet*	
DILUENT: Receiving Water		

CONC	REP	SURVIVAL 0	SURVIVAL 24	SURVIVAL 48	DO (mg/L) 0	DO (mg/L) 24	DO (mg/L) 48	pH (SU) 0	pH (SU) 24	pH (SU) 48	TEMP (°C) 0	TEMP (°C) 24	TEMP (°C) 48	S/C (µmhos/cm) 0	S/C (µmhos/cm) 24	S/C (µmhos/cm) 48	SALINITY (ppt) 0	SALINITY (ppt) 24	SALINITY (ppt) 48
25%	A	10	9	9	7.6	5.3	5.9	7.88	7.85	7.91	24	24	24	39130	39190	39510	25	25	25
	B	10	10	10	7.6	5.4	5.9												
	C	10	10	10	7.6	5.5	5.8												
	D	10	10	10	7.6	5.6	5.8												
50%	A	10	10	10	7.6	5.3	5.8	7.79	7.87	7.95	24	24	24	39980	40080	40410	25	26	26
	B	10	10	10	7.6	4.8	5.5												
	C	10	10	10	7.6	5.0	5.6												
	D	10	10	10	7.6	5.1	5.6												
100%	A	10	8	(E10)	7.7	4.9	(E10)	7.70	7.98	7.99	24	24	24	41510	41760	41910	26	27	27
	B	10	10	9	7.7	4.8	5.4												
	C	10	10	10	7.7	4.6	5.2												
	D	10	9	9 (E10)	7.7	4.8	~~5.1~~												
DATE		5/14/15	5/15/15	5/16/15	05/14/15	05/15/15	5/16												
TIME		1430	1550	1345	1230	1400	1215												
INITIALS		BL	NP	BG	EH	NP	BL												

(E10) beaker knocked over before WQ could be taken BL 5/16 all fish lost

Organism Wet Weights

Study: 25910
Client: Hull
Date/Time/Initials: 05/14/15 1400 NP
Start/End?: Start
Instrument Used: Fisher Accu - 225D

Rep	
1	0.00133
2	0.0028
3	0.00059
4	0.00066
5	0.00209
6	0.00118
7	0.00119
8	0.0003
9	0.00036
10	0.00092
11	0.00053
12	0.0006
13	0.00046
14	0.0005
15	0.00036
16	0.00048
17	0.00055
18	0.00063
19	0.00061
20	0.0004

Mean Weight (g):	0.000827
Test Volume (L):	0.2
Loading Rate(g/L)	0.04135

CETIS Summary Report

Menidia beryllina 48-Hr Acute Survival Test — **EnviroSystems, Inc.**

Batch ID:	18-1545-4190	**Test Type:**	Survival	**Analyst:**	
Start Date:	14 May-15 14:30	**Protocol:**	EPA/821/R-02-012 (2002)	**Diluent:**	Receiving Water
Ending Date:	16 May-15 13:45	**Species:**	Menidia beryllina	**Brine:**	Generic commercial salts
Duration:	47h	**Source:**	ABS - Aquatic Biosystems, CO	**Age:**	8 d

Sample ID:	01-2438-8407	**Code:**	25910	**Client:**	United Water - Hull WWTF
Sample Date:	13 May-15 08:00	**Material:**	Municipal WWTF Effluent	**Project:**	Second Quarter WET Compliance Tes
Receive Date:	13 May-15 11:00	**Source:**	Hull MA WWTF		
Sample Age:	30h (4 °C)	**Station:**	MA0101231		

Comparison Summary

Analysis ID	Endpoint	NOEL	LOEL	TOEL	PMSD	TU	Method
10-5921-3168	Proportion Survived	100	>100	NA	NA	1	Dunn/Bonferroni Test

Point Estimate Summary

Analysis ID	Endpoint	Level	%	95% LCL	95% UCL	TU	Method
20-5143-6811	Proportion Survived	EC10	>100	N/A	N/A	<1	Linear Interpolation (ICPIN)
		EC25	>100	N/A	N/A	<1	

Proportion Survived Summary

C-%	Control Type	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0	Lab Seawater	4	0.908	0.739	1	0.8	1	0.0534	0.107	11.7%	0.0%
0	Receiving Water	4	0.85	0.691	1	0.8	1	0.05	0.1	11.8%	6.42%
6.25		4	0.875	0.477	1	0.5	1	0.125	0.25	28.6%	3.67%
12.5		4	1	1	1	1	1	0	0	0.0%	-10.1%
25		4	0.975	0.895	1	0.9	1	0.025	0.05	5.13%	-7.34%
50		4	1	1	1	1	1	0	0	0.0%	-10.1%
100		3	0.933	0.79	1	0.9	1	0.0333	0.0577	6.19%	-2.75%

Proportion Survived Detail

C-%	Control Type	Rep 1	Rep 2	Rep 3	Rep 4
0	Lab Seawater	1	1	0.8	0.833
0	Receiving Water	1	0.8	0.8	0.8
6.25		1	1	0.5	1
12.5		1	1	1	1
25		0.9	1	1	1
50		1	1	1	1
100			0.9	1	0.9

Menidia beryllina 48-Hr Acute Survival Test — **EnviroSystems, Inc.**

Analysis ID:	10-5921-3168	**Endpoint:**	Proportion Survived	**CETIS Version:**	CETISv1.8.6
Analyzed:	19 May-15 10:42	**Analysis:**	Nonparametric-Control vs Treatments	**Official Results:**	Yes

Sample ID:	01-2438-8407	**Code:**	25910	**Client:**	United Water - Hull WWTF
Sample Date:	13 May-15 08:00	**Material:**	Municipal WWTF Effluent	**Project:**	Second Quarter WET Compliance Tes
Receive Date:	13 May-15 11:00	**Source:**	Hull MA WWTF		
Sample Age:	30h (4 °C)	**Station:**	MA0101231		

Data Transform	Zeta	Alt Hyp	Trials	Seed	NOEL	LOEL	TOEL	TU
Angular (Corrected)	NA	C > T	NA	NA	100	>100	NA	1

Dunn/Bonferroni Test

Control	vs	C-%	Test Stat	Critical	MSD	DF	P-Value	P-Type	Decision(α:5%)
Receiving Water		6.25	-1.6	2.33		6	1.0000	Asymp	Non-Significant Effect
		12.5	-2.53	2.33		6	1.0000	Asymp	Non-Significant Effect
		25	-1.92	2.33		6	1.0000	Asymp	Non-Significant Effect
		50	-2.53	2.33		6	1.0000	Asymp	Non-Significant Effect
		100	-0.842	2.33		5	1.0000	Asymp	Non-Significant Effect

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(α:5%)
Between	0.1701464	0.03402927	5	1.44	0.2606	Non-Significant Effect
Error	0.4018213	0.02363655	17			
Total	0.5719677		22			

Distributional Tests

Attribute	Test	Test Stat	Critical	P-Value	Decision(α:1%)
Variances	Mod Levene Equality of Variance	0.559	4.44	0.7297	Equal Variances
Variances	Levene Equality of Variance	5.33	4.34	0.0040	Unequal Variances
Distribution	Shapiro-Wilk W Normality	0.826	0.88	0.0010	Non-normal Distribution

Proportion Survived Summary

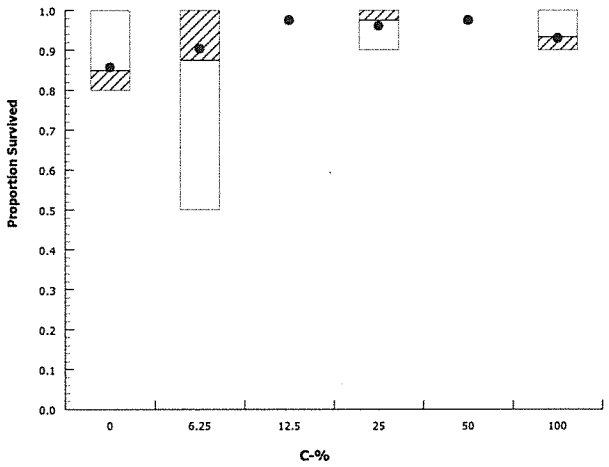
C-%	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Receiving Water	4	0.85	0.691	1	0.8	0.8	1	0.05	11.8%	0.0%
6.25		4	0.875	0.477	1	1	0.5	1	0.125	28.6%	-2.94%
12.5		4	1	1	1	1	1	1	0	0.0%	-17.6%
25		4	0.975	0.895	1	1	0.9	1	0.025	5.13%	-14.7%
50		4	1	1	1	1	1	1	0	0.0%	-17.6%
100		3	0.933	0.79	1	0.9	0.9	1	0.0333	6.19%	-9.8%

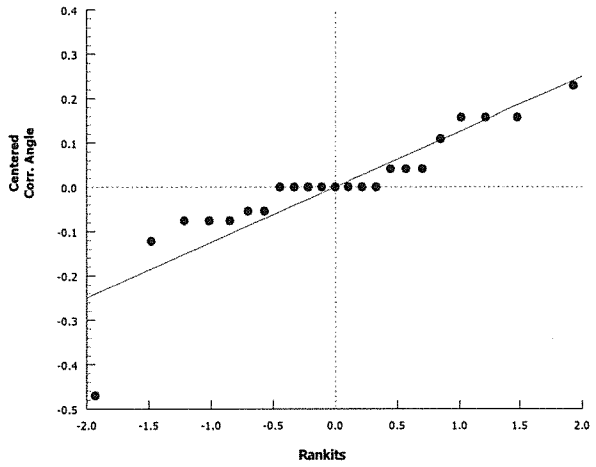
Angular (Corrected) Transformed Summary

C-%	Control Type	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	Receiving Wate	4	1.18	0.941	1.43	1.11	1.11	1.41	0.0762	12.9%	0.0%
6.25		4	1.26	0.757	1.75	1.41	0.785	1.41	0.157	25.0%	-6.08%
12.5		4	1.41	1.41	1.41	1.41	1.41	1.41	0	0.0%	-19.3%
25		4	1.37	1.24	1.5	1.41	1.25	1.41	0.0407	5.94%	-15.9%
50		4	1.41	1.41	1.41	1.41	1.41	1.41	0	0.0%	-19.3%
100		3	1.3	1.07	1.54	1.25	1.25	1.41	0.0543	7.22%	-10.1%

Menidia beryllina 48-Hr Acute Survival Test					EnviroSystems, Inc.
Analysis ID:	10-5921-3168	**Endpoint:**	Proportion Survived	**CETIS Version:**	CETISv1.8.6
Analyzed:	19 May-15 10:42	**Analysis:**	Nonparametric-Control vs Treatments	**Official Results:**	Yes

Graphics





CETIS Analytical Report

Menidia beryllina 48-Hr Acute Survival Test — **EnviroSystems, Inc.**

Analysis ID:	20-5143-6811	**Endpoint:**	Proportion Survived	**CETIS Version:**	CETISv1.8.6
Analyzed:	19 May-15 10:43	**Analysis:**	Linear Interpolation (ICPIN)	**Official Results:**	Yes

Sample ID:	01-2438-8407	**Code:**	25910	**Client:**	United Water - Hull WWTF
Sample Date:	13 May-15 08:00	**Material:**	Municipal WWTF Effluent	**Project:**	Second Quarter WET Compliance Tes
Receive Date:	13 May-15 11:00	**Source:**	Hull MA WWTF		
Sample Age:	30h (4 °C)	**Station:**	MA0101231		

Linear Interpolation Options

X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Log(X+1)	Linear	376325	200	Yes	Two-Point Interpolation

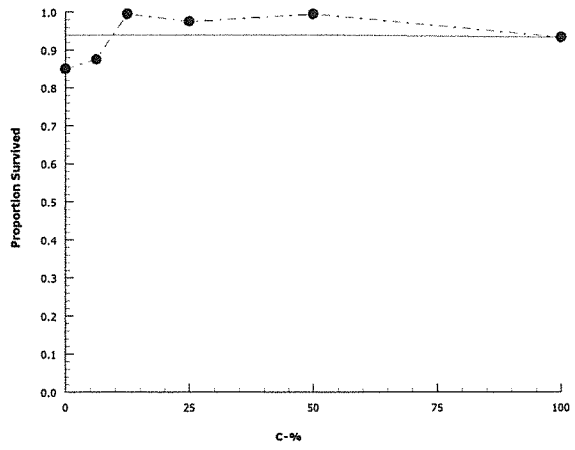
Point Estimates

Level	%	95% LCL	95% UCL	TU	95% LCL	95% UCL
EC10	>100	N/A	N/A	<1	NA	NA
EC25	>100	N/A	N/A	<1	NA	NA

Proportion Survived Summary — Calculated Variate(A/B)

C-%	Control Type	Count	Mean	Min	Max	Std Err	Std Dev	CV%	%Effect	A	B
0	Receiving Water	4	0.85	0.8	1	0.05	0.1	11.8%	0.0%	34	40
6.25		4	0.875	0.5	1	0.125	0.25	28.6%	-2.94%	35	40
12.5		4	1	1	1	0	0	0.0%	-17.6%	40	40
25		4	0.975	0.9	1	0.025	0.05	5.13%	-14.7%	39	40
50		4	1	1	1	0	0	0.0%	-17.6%	40	40
100		3	0.933	0.9	1	0.0333	0.0577	6.19%	-9.8%	28	30

Graphics



07M6#BS051315

1300 Blue Spruce Drive, Suite C
Fort Collins, Colorado 80524

Toll Free: 800/331-5916
Tel: 970/484-5091 Fax:970/484-2514

ORGANISM HISTORY

DATE: 5/12/2015

SPECIES: *Menidia beryllina*

AGE: 6 day

LIFE STAGE: Juvenile

HATCH DATE: 5/6/2015

BEGAN FEEDING: Immediately

FOOD: Rotifers, *Artemia* sp.

Water Chemistry Record:	**Current**	**Range**
TEMPERATURE:	25°C	23-26 °C
SALINITY/CONDUCTIVITY:	25 ppt	24-26 ppt
TOTAL HARDNESS (as $CaCO_3$):	--	--
TOTAL ALKALINITY (as $CaCO_3$):	150 mg/l	150-200 mg/l
pH:	8.18	7.70-8.20

Comments:

Facility Supervisor

RECORD OF METERS USED

STUDY: 25910	CLIENT: United Water - Hull, MA WWTF		
Exposure (Hours)			
	0	24	48
Water Quality Station #	2	2	1
Initials / Date	EH 05/14/15	5/15/15 NP	~~JG~~ BL 05/16/15 (FS) JG 5/16

Water Quality Station #1		Water Quality Station #2		COMMENTS
DO meter #	24	DO meter #	23	
DO probe #	94	DO probe #	93	
pH meter #	1097	pH meter #	470	
pH probe #	134	pH probe #	132	
S/C meter #	YSI30E	S/C meter #	YSI30E	
S/C probe #	↓	S/C probe #	↓	
Salinity meter #	↓	Salinity meter #	↓	

PREPARATION OF DILUTIONS

Diluent: Receiving Water (RW)	Day: 0 Sample: E_0, D_0	
Concentration %	Vol. Eff.(mls)	Final Vol.(mls)
Lab	0	800
RW	0	↓
6.25%	50	
12.5%	100	
25%	200	
50%	400	
100%	800	
INITIALS:	EH	
TIME:	1145	
DATE:	05/14/15	

Report No: 25910 SDG:
Project: Hull

Sample ID: Effluent Start
Matrix: Water
Sampled: 05/13/15 0800

Parameter		Result	Quant Limit	Units	Date Prepared	Date of Analysis	INIT/Method/Reference
Total solids	25910-005	5000	10	mg/L	05/18/15 1430	05/27/15 0945	BG /SM2540B
Total suspended solids	25910-005	23	2	mg/L	05/14/15 0950	05/18/15 1500	BG /SM 2540D
Total organic carbon	25910-003	11	0.8	mg/L	05/14/15	05/14/15	MG /SM 5310 C
Ammonia-N	25910-004	16	0.1	mg/L as N	05/15/15 1034	05/15/15 1034	MG /SM 4500-NH3 G
Aluminum, total	25910-002	0.09	0.02	mg/L	05/20/15	05/20/15	JLH/EPA 200.8
Cadmium, total	25910-002	ND	0.0005	mg/L	05/20/15	05/20/15	JLH/EPA 200.8
Calcium, total	25910-002	98	0.05	mg/L	05/20/15	05/20/15	JLH/EPA 200.8
Chromium, total	25910-002	ND	0.002	mg/L	05/20/15	05/20/15	JLH/EPA 200.8
Copper, total	25910-002	0.37	0.002	mg/L	05/20/15	05/20/15	JLH/EPA 200.8
Lead, total	25910-002	0.001	0.0005	mg/L	05/20/15	05/20/15	JLH/EPA 200.8
Magnesium, total	25910-002	210	0.05	mg/L	05/20/15	05/20/15	JLH/EPA 200.8
Nickel, total	25910-002	0.003	0.002	mg/L	05/20/15	05/20/15	JLH/EPA 200.8
Zinc, total	25910-002	0.14	0.002	mg/L	05/20/15	05/20/15	JLH/EPA 200.8

Sample ID: Receiving Water Start
Matrix: Water
Sampled: 05/13/15 0700

Parameter		Result	Quant Limit	Units	Date Prepared	Date of Analysis	INIT/Method/Reference
Total solids	25910-010	35000	100	mg/L	05/18/15 1430	05/27/15 0945	BG /SM2540B
Total suspended solids	25910-010	6.3	1	mg/L	05/14/15 0950	05/18/15 1500	BG /SM 2540D
Total organic carbon	25910-008	0.7	0.4	mg/L	05/14/15	05/14/15	MG /SM 5310 C
Ammonia-N	25910-009	ND	0.1	mg/L as N	05/15/15 1034	05/15/15 1034	MG /SM 4500-NH3 G
Aluminum, total	25910-007	0.099	0.02	mg/L	05/20/15	05/20/15	JLH/EPA 200.8
Cadmium, total	25910-007	ND	0.0005	mg/L	05/20/15	05/20/15	JLH/EPA 200.8
Calcium, total	25910-007	360	0.05	mg/L	05/20/15	05/20/15	JLH/EPA 200.8
Chromium, total	25910-007	ND	0.002	mg/L	05/20/15	05/20/15	JLH/EPA 200.8
Copper, total	25910-007	0.006	0.002	mg/L	05/20/15	05/20/15	JLH/EPA 200.8
Lead, total	25910-007	ND	0.0005	mg/L	05/20/15	05/20/15	JLH/EPA 200.8
Magnesium, total	25910-007	1000	0.05	mg/L	05/20/15	05/20/15	JLH/EPA 200.8
Nickel, total	25910-007	ND	0.002	mg/L	05/20/15	05/20/15	JLH/EPA 200.8
Zinc, total	25910-007	0.007	0.002	mg/L	05/20/15	05/20/15	JLH/EPA 200.8

Notes:

ND = Not Detected

SAMPLE RECEIPT AND CONDITION DOCUMENTATION

STUDY NO:	25910		
SDG No:			
Project:	Hull		
Delivered via:	ESI		
Date and Time Received:	05/13/15 1100	Date and TIme Logged into Lab:	05/13/15 1400
Recieved By:	DW EH	Logged into Lab by:	EH EH
Air bill / Way bill:	No	Air bill included in folder if received?	NA
Cooler on ice/packs:	Yes	Custody Seals present?	NA
Cooler Blank Temp (C) at arrival:	4.2	Custody Seals intact?	NA
Number of COC Pages:	1		
COC Serial Number(s):	A1011783		
COC Complete:	Yes	Does the info on the COC match the samples?	Yes
Sampled Date:	Yes	Were samples received within holding time?	Yes
Field ID complete:	Yes	Were all samples properly labeled?	Yes
Sampled Time:	Yes	Were proper sample containers used?	Yes
Analysis request:	Yes	Were samples received intact? (none broken or leaking)	Yes
COC Signed and dated:	Yes	Were sample volumes sufficient for requested analysis?	Yes
Were all samples received?	Yes	Were VOC vials free of headspace?	NA
Client notification/authorization:	Not required		

Field ID	Lab ID	Mx	Analysis Requested	Bottle	Req'd Pres'n	Verified Pres'n
Effluent Start	25910-001	W	MB48AD StartSample	1x3750 P	4 C	
Effluent Start	25910-002	W	Total Metals Cd,Cr,Ni,Pb,Cu,Zn,Al,Ca,Mg;	250 P	HNO3	Yes
Effluent Start	25910-003	W	TOC	1x40 G	H2SO4	Yes
Effluent Start	25910-004	W	NH3;	125 P	H2SO4	Yes
Effluent Start	25910-005	W	TS,TSS	1000 P	4 C	
Receiving Water Start	25910-006	W	MB48AD StartDiluent	2x3750 P	4 C	
Receiving Water Start	25910-007	W	Total Metals Cd,Cr,Ni,Pb,Cu,Zn,Al,Ca,Mg;	250 P	HNO3	Yes
Receiving Water Start	25910-008	W	TOC	1x40 G	H2SO4	Yes
Receiving Water Start	25910-009	W	NH3;	125 P	H2SO4	Yes
Receiving Water Start	25910-010	W	TS,TSS	1000 P	4 C	

Notes and qualifications:

See COC

ESI EnviroSystems, Inc.
1 Lafayette Road
Hampton, NH 03842

Voice: 603-926-3345
FAX: 603-926-3521

ESI Job No: 25910

CHAIN OF CUSTODY DOCUMENTATION

Client: Hull	Contact: Aram Varjabedian	Project Name: Hull WWTF	
Report to: Aram Varjabedian	Address: 1111 Nantasket Avenue	Project Number: P0036	Task: 0001
Invoice to: Aram Varjabedian	Address: Hull, MA 02045	Project Manager: Aram Varjabedian	
Voice: 781-925-0906	Fax: 781-925-3056	email:	P.O.No: ' Quote No:41181

Protocol: NPDES

Lab Number (assigned by lab)	Your Field ID: (must agree with container)	Date Sampled	Time Sampled	Sampled By	Grab or composite (G/C)	Container No	Container Size (mL)	Container Type (P/G/T)	Field Preservation	Matrix S=Solid W=Water	Filter N=Not needed F=Done in field L=Lab to do	Analyses Requested\ Special Instructions:
001	Effluent Start	5/12-13/15	8A-8A		C	1	3750	P	4 C	Water	N	MB48AD StartSample
002	Effluent Start	5/12-13/15	8A-8A		C	1	250	P	HNO3	Water	N	Total Metals Cd,Cr,Ni,Pb,Cu,Zn,Al,Ca,Mg;
003	Effluent Start	5/12-13/15	8A-8A		C	1	40	G	H2SO4	Water	N	TOC
004	Effluent Start	5/12-13/15	8A-8A		C	1	125	P	H2SO4	Water	N	NH3;
005	Effluent Start	5/12-13/15	8A-8A		C	1	1000	P	4 C	Water	N	TS,TSS
006	Receiving Water Start	5/13/15	7A		G	2	3750	P	4 C	Water	N	MB48AD StartDiluent
007	Receiving Water Start	5/13/15	7A		G	1	250	P	HNO3	Water	N	Total Metals Cd,Cr,Ni,Pb,Cu,Zn,Al,Ca,Mg;
008	Receiving Water Start	5/13/15	7A		G	1	40	G	H2SO4	Water	N	TOC
009	Receiving Water Start	5/13/15	7A		G	1	125	P	H2SO4	Water	N	NH3;
010	Receiving Water Start	5/13/15	7A		G	1	1000	P	4 C	Water	N	TS,TSS

Relinquished By: [signature] Date: 5/13/15 Time: 11:00

Received By: [signature] Date: 5/13/15 Time: 11:00

Relinquished By: Date: Time:

Received at Lab By: Date: Time:

Comments: 4.2°C

ERR

COC Number: A1011783

Sample Delivery Group No: April 2015

Page of

Assay Review Checklist

DATE IN: 5/13/15
DATE DUE: 6/10/15

STUDY#: 25910
CLIENT: United Water
PROJECT: Hull
ASSAY: MB48AD

Project Paperwork Check for Completeness	Date	Initials	Comments
Day 0	5/14/15	BC	
Day 1	5/15	NP	
Day 2	5/16/15	BG	ARC Day 1
Day 3			
Day 4			
Day 5			
Day 6			
Day 7			
Day 8			

Analyst Data Review	Date	Initials	Comments
Chains of Custody Complete	5/18/15	UB	
Sample Receipt Complete	↓	↓	
Organism Culture Sheet(s)			
Bench Sheets Complete (dates, times, initials, etc...)			
Water Quality Data Complete			
TRC Values & Bottle Numbers	↓		
Daphnid Calculations Complete	N/A		
Weights Reported	5/18/15		
Assay Acceptability Review	↓	↓	

Technical Report Review	Date	Initials	Comments
Statistical Analysis Complete	5/19/15	NR	MB-12D had 12 orgs from start - 100A removed from stats - stats not run using std EPA dec. tree b/c of # of groups/reps selected at spec. level. Calc'd using Dunn/Bonf + linear interpolation (IC25) instead.
Statistical Analysis Reviewed	5/19/15	UB	
Data Acceptability Review	5/19/15	NR	
Supporting Chemistry Report	6/2/15	NR	
Draft Report	5/19/15	UB	
QA Audit/Review Complete	5/26/15	NR	
Final Report Reviewed	EB NR 6/2/15		
Final Report Printed - PDF	6/2/15	NR	
Executive Summary / Chems Sent			
Report E-mailed / Faxed	6/2/15	NR	
Report Logged Out / Invoice Sent	↓	↓	
Report Scanned to Archive	↓		

P:\GENERAL PROJECTS\FORMS\LABFORMS\$ Assay Review Checklist.wpd